



SEQUENCE LISTING

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#4

<120> COMBINATORIAL POLYKETIDE LIBRARIES
PRODUCED USING A MODULAR PKS GENE CLUSTER AS SCAFFOLD

<130> 30062-20005.02

<140> 09/859,854
<141> 2001-05-16

<150> PCT/US98/08792
<151> 1998-04-30

<150> 60/076,919
<151> 1998-03-05

<150> 08/846,247
<151> 1997-04-30

<150> 08/486,645
<151> 1995-06-07

<150> 08/238,811
<151> 1994-05-06

<160> 44

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<223> Module 1 - A BamHI site engineered for the 5'
boundary of the acyltransferase domain.

<400> 1
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24

<210> 2
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<213> Artificial Sequence

<220>
<223> Module 1 - A PstI site engineered for introduction
between the acyltransferase and reductive domains.

<400> 2
cgcgtctggc tgcagccgaa gccg 24

<210> 3
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 1 - A XbaI site engineered for introduction at the 3' end of the reductive domain.

<400> 3
gcgcgggtga gatctaagcc ggcc 24

<210> 4
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<213> Artificial Sequence

<220>
<223> Module 2 - A BamHI site engineered for the 5' boundary of the acyltransferase domain.

<400> 4
tccgacggtg gatccgtgtt cgtc 24

<210> 5
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 2 - A PstI site engineered for introduction between the acyltransferase and reductive domains.

<400> 5
cggttctggc tgcagccgga ccgc 24

<210> 6
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 2 - A XbaI site engineered for introduction at the 3' end of the reductive domain.

<400> 6
gtcggccaga gatctcgaga ggca 24

<210> 7
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 3 - A BamHI site engineered for the 5'
boundary of the acyltransferase domain.

<400> 7
gacgggcgatccgtctt cctg 24

<210> 8
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 3 - A PstI site engineered for introduction
between the acyltransferase and reductive domains.

<400> 8
cgctactggc tgcaagccgc cgca 24

<210> 9
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 3 - A XbaI site engineered for introduction
at the 3' end of the reductive domain.

<400> 9
ctcggcaaca gatctgagcg gcca 24

<210> 10
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 4 - A BamHI site engineered for the 5'
boundary of the acyltransferase domain.

<400> 10
gcggccgcgatccgtcct ggtc 24

<210> 11
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 4 - A PstI site engineered for introduction
between the acyltransferase and reductive domains.

<400> 11
cgcttctggc tgcaagccgca ccgg 24

<210> 12
<211> 24

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<212> DNA
<213> Artificial Sequence

<220>
<223> Module 4 - A XbaI site engineered for introduction
      at the 3' end of the reductive domain.

<400> 12
      ctcggccaga gatctcaagg cggg          24

<210> 13
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 5 - A BamHI site engineered for the 5'
      boundary of the acyltransferase domain.

<400> 13
      actcgccgca gatccgcgtt ggtg          24

<210> 14
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 5 - A PstI site engineered for introduction
      between the acyltransferase and reductive domains.

<400> 14
      cggtaactggc tgcagatccc cacc          24

<210> 15
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 5 - A XbaI site engineered for introduction
      at the 3' end of the reductive domain.

<400> 15
      gacccgctca gatctcgaaa ggag          24

<210> 16
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 6 - A BamHI site engineered for the 5'
      boundary of the acyltransferase domain.

<400> 16
      tccggccggcg gatccgtttt cgtc          24

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<210> 17
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 6 - A PstI site engineered for introduction between the acyltransferase and reductive domains.

<400> 17
cggtactggc tgcaagccgga ggtg 24

<210> 18
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Module 6 - A XbaI site engineered for introduction at the 3' end of the reductive domain.

<400> 18
gacgtggcga gatctccggg ggtg 24

<210> 19
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> A PstI site that is in-frame and upstream of XbaI in pUC19 that generates this junction at the 5' end of the cassette.

<400> 19
ctgcaggatcg actcttagcct ggt 23

<210> 20
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapAT2 (forward) Primer pairs used for PCR amplification of rapamycin PKS cassettes.

<400> 20
tttagatctg tggcgtctt cccgggt 27

<210> 21
<211> 36
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapAT2 (reverse) Primer pairs used for PCR

amplification of rapamycin PKS cassettes.

<400> 21		
tttctgcagc cagtaccgct ggtgctggaa ggcgtt		36
<210> 22		
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<213> Artificial Sequence		
<220>		
<223> Module rapAT14 (forward) Primer pairs used for PCR		
amplification of rapamycin PKS cassettes.		
<400> 22		
tttggatccg ctttcctgtt cgacgggcaa ggc		33
<210> 23		
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<223> Module rapAT14 (reverse) Primer pairs used for PCR		
amplification of rapamycin PKS cassettes.		
<400> 23		
tttctgcagc cagtaggact ggtgctggaa cgg		33
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<213> Artificial Sequence		
<220>		
<223> Module rapKR2 (forward) Primer pairs used for PCR		
amplification of rapamycin PKS cassettes.		
<400> 24		
tttctgcagg agggcacgga ccgggcgact gcgggt		36
<210> 25		
<211> 36		
<212> DNA		
<213> Artificial Sequence		
<220>		
<223> Module rapKR2 (reverse) Primer pairs used for PCR		
amplification of rapamycin PKS cassettes.		
<400> 25		
ttttctagaa ccggcggcag cggcccggccg agcaat		36
<210> 26		
<211> 26		
<212> DNA		
<213> Artificial Sequence		

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<220>
<223> Module rapDH/KR4 (forward) Primer pairs used for
      PCR amplification of rapamycin PKS cassettes.

<400> 26
ttctgcagag cgtggaccgg gcggct                                26

<210> 27
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapDH/KR4 (reverse) Primer pairs used for
      PCR amplification of rapamycin PKS cassettes.

<400> 27
ttttcttagag tcacccgttag aggccggccct                                30

<210> 28
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapDH/ER/KR1 (forward) Primer pairs used
      for PCR amplification of rapamycin PKS cassettes.

<400> 28
tttctgcagg gcgtggaccg ggcggctgcc                                30

<210> 29
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapDH/ER/KR1 (reverse) Primer pairs used
      for PCR amplification of rapamycin PKS cassettes.

<400> 29
tttctcgagc accacgccccg cagcctcacc                                30

<210> 30
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapDH/ER/KR1 (forward) Primer pairs used
      for PCR amplification of rapamycin PKS cassettes.

<400> 30
tttctcgagg tcggtccgga ggtccaggat                                30

<210> 31

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<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Module rapDH/ER/KR1 (reverse) Primer pairs used
for PCR amplification of rapamycin PKS cassettes.

<400> 31
ttttctagaa tcacccgttag aaggcagccccg 30

<210> 32
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> The junctions at which the PstI and XbaI sites
were introduced into DEBS.

<400> 32
gagcccccagc ggtactggct gcag 24

<210> 33
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> The junctions at which the PstI and XbaI sites
were introduced into DEBS.

<400> 33
tcttagagcgg tgcaggcggc cccg 24

<210> 34
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> A primer at which the eryKR6 domain was PCR
amplified.

<400> 34
tttggatccg ttttcgtctt cccaggtcag 30

<210> 35
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> A primer at which the eryKR6 domain was PCR
amplified.

<400> 35

tttctgcagc cagtaccgct ggggctcgaa

30

<210> 36
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> A primer at which the eryKR6 domain was PCR amplified.

<400> 36
ttttcttagag cggtgcaggc ggccccggcg

30

<210> 37
<211> 29
<212> DNA
<213> Artificial Sequence

<220>
<223> A primer at which the eryKR6 domain was PCR amplified.

<400> 37
aaaatgcattc tatgaattcc ctccggcca

29

<210> 38
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> The resulting PstI and XbaI junctions engineered into DEBS.

<400> 38
gaacaccaggc gcttctggct gcag

24

<210> 39
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> The resulting PstI and XbaI junctions engineered into DEBS.

<400> 39
tctagagagacc ggctcgccgg tcgg

24

<210> 40
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Resulting engineered DEBS/rapAT2 junction.

<400> 40
agtgcctccg acgggtggatc t 21

<210> 41
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Resulting engineered DEBS/rapAT2 junction.

<400> 41
ctgcagccgg accgcaccac ccct 24

<210> 42
<211> 47
<212> DNA
<213> Artificial Sequence

<220>
<223> A synthetic oligonucleotide linker designed to allow complete excision of reductive cycle domains - designed to generate PstI- and XbaI-compatible ends upon hybridization.

<400> 42
gccggaccgc accacccctc gtgacggaga accggagacg gagagct 47

<210> 43
<211> 55
<212> DNA
<213> Artificial Sequence

<220>
<223> A synthetic oligonucleotide linker designed to allow complete excision of reductive cycle domains - designed to generate PstI- and XbaI-compatible ends upon hybridization.

<400> 43
ctagagctct ccgtctccgg ttctccgtca cgaggggtgg tgcggtccgg ctgca 55

<210> 44
<211> 12
<212> DNA
<213> Artificial Sequence

<220>
<223> The fusion between the two residues L3455 of DEBS1 and Q2891 of DEBS3.

<400> 44
ctcactagtc ag 12